CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA
ACEPHATE

Chemical Code # 001685, Tolerance # 00108
SB 950 # 125

October 1, 1986
Revised 2/5/87; 1/25/88; 7/2/88; 11/7/88; 4/6/90; 5/5/93, 2/8/02 and 6/7/02

I. DATA GAP STATUS

Combined, rat:	No data gap, no adverse effect
Chronic toxicity, rat:	See Combined, rat
Chronic toxicity, dog:	No data gap, possible adverse effect
Oncogenicity, rat:	See Combined, rat
Oncogenicity, mouse:	No data gap, possible adverse effect
Reproduction, rat:	No data gap, no adverse effect
Teratology, rat:	No data gap, no adverse effect
Teratology, rabbit:	No data gap, no adverse effect
Gene mutation:	No data gap, possible adverse effect
Chromosome mutation:	No data gap, possible adverse effect
DNA damage:	No data gap, possible adverse effect
Neurotoxicity:	No data gap, possible adverse effect (rats)

Toxicology one-liners are attached.
** Indicates an acceptable study.
Bold face indicates a possible adverse effect.
FILE NAME: T020607
Revised by M. Silva, 11/88 and 4/90; Kellner, 5/5/93 and Gee, 2/8/02 and 6/7/02
All relevant records indexed as of 2/8/02 were included in this summary.
These pages contain summaries only. Each individual worksheet may contain additional effects.

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

COMBINED, RAT

** 139, 144 037723, 037728 "Lifetime Oral Toxicity/Carcinogenicity Study with Technical RE-12420 (Orthene) in Rats." (Bio/dynamics, 6/30/81). Acephate (92.4%, lot SX-992) fed in the diet at 0, 5, 50 or 700 ppm (0.24/0.31, 2.44/3.06 or 38.2/47.2 mg/Kg, M/F), 80/sex/group; significantly lower body weight in males at high dose; consistent cholinesterase inhibition at high dose and to a lesser extent at low and mid dose levels; systemic NOEL = 5.0 ppm (based on brain cholinesterase). McGee evaluation (4/28/86) was unacceptable but upgradeable. Davis evaluation (1/5/87) of supplemental data and Chevron response was complete and ACCEPTABLE.

No EPA one-liner available.

067 973192 Interim report for record 037723.

067 973187 Supplement to record 037723--photomicrographs.

136 026943 Supplement to 037723--discussion of amendments to report.

146 037732 Supplement to 037723--rationale for amendments to report.

067 971388 Less complete version of study identified as record 037723. Reviewed by J.Wong, 5/22/85, with insufficient information for evaluation.

160 050280 (Bio/Dynamics, 3/20/78) Supplement to 108-139 to 144 and 108-46, 037723-8 --Diet analysis data. Samples from cage hoppers after 3-4 days were 93.1% of nominal for the 5 ppm level, 81.8% for the 50 ppm level, and 80.7% for the 700 ppm level. Problems in the diet analysis included acephate found in the negative control samples between 5/5/78 and 7/3/78, corrections needed in the calculations on most pages, and missing chromatograms. With this addendum and the information from the Chevron response of 11/24/86, the study is complete and ACCEPTABLE. Davis 1/5/87.

161 Rebuttal of 11/20/86 to DPR review of 037723-8.

CHRONIC TOXICITY, RAT

015 973190 Invalid IBT study, 1/29/73.

CHRONIC TOXICITY, DOG

**** 108-238 096115** "One-Year Oral Toxicity Study in Dogs with Chevron Acephate Technical." (D.W. Dalgard, Hazleton Washington, HWA 2107-165, 1/25/91). Acephate technical, purity of 99.9%, was administered in the feed at concentrations of 0, 10, 120 (reduced from 200 ppm during week 2 of study), or 800 ppm to 5 Beagle dogs/sex/group for 1 year. High-dose groups had lower red cell mass indices, elevated APTT and increased liver weights. Liver pathology (perivascular pleocellular infiltrate and pigment in the reticuloendothelial cells) was noted in most high-dose animals and one mid-dose male (NOEL = 10 ppm/day). Significant RBC cholinesterase (ChE) inhibition for mid- and high dose groups was reported in addition to brain ChE inhibition for mid- and high dose females and all dose levels for males. ChE NOEL (females) = 10 ppm; males < 10 ppm. **Possible Adverse Effect:** Significant brain ChE inhibition (no NOEL in males). ACCEPTABLE. Kishiyama, Kellner and Aldous, 4/30/93.

108-237 095950. This submission was an adverse effect disclosure for 096115. Adverse Effect consisted of a significant (16%) inhibition of brain ChE in low dose (10 ppm) males by the end of the study. No worksheet. Kishiyama and Kellner, 4/30/93.

108-225 85614 "Four-Week Pilot Oral Toxicity Study in Dogs with Chevron Acephate Technical" (Hazleton Laboratories, Chevron Report #2107-164). Levels of acephate of 250 and 500 ppm in the feed resulted in brain ChE levels of 6.2 and 5.0 mmol/g, respectively. A brain ChE NOEL was established at 20 ppm. No worksheet. Kellner, 5/6/93.

015 973189 "Two Year Chronic Oral Toxicity Study with RE 12420 in Beagle Dogs." (IBT, No. C-8732, 12/28/72) Acephate (87 to 94 %, < 0.5 % methamidophos content), lots SX-257, 1st six months and SX-357, final 18 months was fed in the diet at 0, 10, 30 or 100 ppm for two years with 4/sex/group. There was a decrease in RBC cholinesterase in both sexes at the high dose level. No adverse effects reported. UNACCEPTABLE (dose selection not adequately justified - high dose may not be sufficient, no analysis of diet for actual content, no ophthalmological exam, inadequate presentation of histopathology). Document 108-169, Record 61136, contains two validation reports including many variations between the raw data and the report and also identifies data not recorded. Not upgradeable. Wong, 5/13/85 and Gee, 1/5/88.

EPA one-liner: NOEL $\geq$ 100 ppm (HDT) for systemic toxicity; cholinesterase activity NOEL = 30 ppm; core grade--minimum.

161 Rebuttal of 11/20/86 to DPR review of 973189.

169 061136 Supplemental to 973189, two validation reports including variations between raw data and the report. Also, stability in dog diet over 7 days at room temperature. Gee, 1/5/88.

012 046560 One year interim report for study identified as record #973189.

170 061137 "90-Day Subacute Oral Toxicity Study with Orthene In Beagle Dogs." (IBT, no. C9527, 8-24-71) Range finding study for record number 973189, volume 108-015. Orthene,

SX-284, was administered to beagles at dietary levels of 0, 10, 30 or 100 ppm, 4/sex/group for 90 days. No abnormalities were noted in body weight, food consumption, behavior, clinical studies, necropsy or histopathology except for up to 60% RBC ChE inhibition at the high dose. Dogs were housed 4/kennel, sac 2/sex/group at 90 days, the other 2 were allowed to recover for a 40 day period. EPA has determined the study is "invalid". Shimer, 11/10/87 and Gee, 12/30/87.

165 057929 Validation report of 061137 prepared by F. X. Kamienski of Chevron. A number of discrepancies between the raw data and the report are pointed including the fact that the hematology, clinical chemistry and urinalysis data are from the two-year study, not the range-finding study. Stability, chemical analyses and corrections are contained in the appendices. Gee, 12/30/87.

ONCOGENICITY, RAT

085 973195 "Oral Toxicity/Carcinogenicity Study in Technical RE 12420 in Rats." (Bio/dynamics, 5/14/79, 77-1870). Acephate, lot 016-SFO-8847-8600, SX941 was fed to 70/sex/group at 0, 10, 50 or 250/350 ppm, Sprague-Dawley rats. The two-year study was terminated after 190 days due to the discovery of an impurity in the test article - the impurity was not identified. The ophthalmoscopic exam at three months was negative. UNACCEPTABLE, not upgradeable. Wong, 5/16/85.

ONCOGENICITY, MOUSE

**** 145, 204 037729, 069074** "Orthene Technical (RE-12420) Lifetime Oral Carcinogenicity Study in Mice," (IRDC, 2/24/82). Acephate (purity = 92.7, 92.1%; lot no. SX-1032) was fed in the diet to CD1 mice for 104 weeks at 0 (vehicle = chow), 50, 250 or 1000 ppm (7, 36 or 146 mg/kg/day) for males; 8, 42, or 167 mg/kg/day for females) with 75/sex/group). **Possible adverse effect.** Nominal NOEL = 50 ppm (decreased body weight at mid and high doses; hepatocellular carcinoma, adenoma and hyperplasia were observed in females at the high dose; other dose-related non-neoplastic changes in males and females were observed primarily at mid- and high-doses; microscopic lung changes were observed at all dose levels in both sexes but were not well defined ("pigmented alveolar macrophages," "eosinophilic foreign bodies")). Originally reviewed as unacceptable by McGee, 4/29/86 (no individual data on food consumption; no individual clinical observations; no statistical analysis of tumor data) and not upgradeable, based on lung findings at all treatment levels. In view of the uncertain nature of the lesions and the consideration of this study as an oncogenicity study, it may be upgradeable with submission of the missing data. DPR has received and reviewed the requested information (204 069674), and the study is upgraded to ACCEPTABLE**.** M. Silva, 10/28/88.

EPA one-liner (Partial excerpt): (NOEL not indicated), Increased incidence of hepatocellular carcinomas and hyperplastic nodules in females at high dose level,

dose-related non-neoplastic liver and lung injuries in male and females, decreased weight gain at

the high dose level in male and female; core grade--minimum.

172 061139 Addendum to 37729 - Diet analysis data - duplicate of Reference 2 of 145 037729. A letter at the beginning of the document, dated August 20, 1987, indicated that the data on food consumption and individual clinical observations would be submitted to DPR in September, 1987. Gee, 1/6/88.

085 973194 Interim report of record 037729. (Reviewed by J. Wong, 5/16/85, as unacceptable with a possible adverse oncogenic and/or chronic effect.)

067 973193 Interim report for record 037729.

128 016928 Supplement to record 037729 -- discussion of hepatocarcinomas in female mice.

128 016929 Historical histopathology data for record 037729.

161 Rebuttal of 11/20/86 to DPR review of 037729.

161 050991 Homogeneity of diet mixing for 037729.

Letter of 5/5/88. Chevron has committed to supply individual data as requested.

015 973191 Invalid IBT study, 3/7/73.

REPRODUCTION, RAT

148 037738 "Effect of Technical Re-12420 (Orthene) on Reproductive Function of Multiple Generations in the Rat." (Huntingdon, 4/18/83). Acephate technical (92.8%, SX-1032) fed in the diet at 0, 50, 150 or 500 ppm for a three generation, two litter/generation study with CrL:cobs CD(SD)BR rats, 12 males and 24 females per group. There was reduced fertility, especially in males, reduced pup viability. Parental (maternal) MTD = 500 ppm. Fertility and viability NOEL not determined due to fact that noted effects had a similar frequency at low and high doses but not at mid dose. UNACCEPTABLE (inadequate number of gravid animals per group, incomplete histopathology, other studies conducted in same animal rooms, no standardization of litter size, no historical control data presented). Not upgradeable. A repeat study following guidelines is recommended. McGee, 8/1/86.

No EPA one-liner.

110 973202 Supplement to record 037738--Statistical analysis report.

110 973204 Less complete version of record 037738 (Reviewed by J. Wong, 5/22/85, as unacceptable due to insufficient but with a possible adverse effect identified in the report as

submitted by registrant).

** 182 060979 "Two-Generation (Two Litter) Reproduction Study in Rats with Chevron Acephate Technical." (Argus Research Laboratories: 303005, 4-3-87) Chevron acephate technical, 98.5%, was fed in the diet to Crl:COBS CD (SD) BR rats, 30/sex/group, at 0, 25, 50 or 500 ppm for two generations, 2 litters/generation, and one litter in the third generation. Parental NOEL = 50 ppm (soft/liquid feces), Reproductive NOEL = 50 ppm (reduced litter size and postnatal survival). This study was conducted primarily to address the effects reported in an earlier study (DPR # 37738) in which a NOEL was not established. This present study does demonstrate a NOEL for both viability and fertility. ACCEPTABLE. Shimer, 11/24/87, J. Gee, 12/30/87.

No EPA one-liner.

209 072152 "Two-Generation (Two Litter) Reproduction Study in Rats with Chevron Acephate Technical." This volume contains a final revision of the definitive rat reproduction study (182 060979). Two changes were made: 1. post natal survival at 25 ppm for first generation F1a litter was changed to not be statistically lower than the value for the control. 2. # of pups dying Days 1-4 at 500 ppm for first generation F1a litter was changed to not be statistically significant when compared to control. The data were accompanied by a summary letter. The changes did not alter the outcome of the study, nor did it alter its acceptability. D. Shimer & M. Silva, 3/30/90.

014 973205 Invalid IBT study, 1/10/73.

Summary: The study conducted at Huntingdon, DPR Record #037738, identified a possible adverse reproductive effect on fertility, especially in males, and decreased pup viability at 50 and 500 ppm. These effects were not confirmed in the later study (DPR Record #060979) at 50 ppm with reproductive effects seen only at 500 ppm in the presence of parental effects. The collective data are adequate to fulfill the requirement with the reproductive NOEL at 50 ppm and no effect seen without some parental effects. The overall conclusion is that acephate does not cause adverse reproductive effects. Gee, 1/5/88.

REPRODUCTION, CHICKEN

014 973207 Invalid IBT study on chicken; not a SB950 test species.

111 973081 Reference to record #973207.

TERATOLOGY, RAT

** 219, 230 074315, 088434 "Oral Teratogenicity and Developmental Toxicity Study in Rats with Chevron Acephate Technical," (Argus Research Laboratories, Study No. 303-008, 2-13-89). Acephate technical (Lot #: SX-1725; purity = 99.5%, or Lot #: SX-1102; purity = 98.7%) was administered to mated Crl:CD®(SD)BR rats (25/dose) by gavage on days 6 - 15 of presumed gestation

(presence of vaginal sperm or copulatory plug = day 0 of gestation) at 0, 5, 20 or 75 mg/kg/day. Maternal NOEL = 5 mg/kg (increase in tremors, decrease in motor activity, body weight and food consumption). Developmental NOEL = 20 mg/kg (decreased fetal body weight, and delay in ossification of hindpaw phalanges--historical controls for malformations and alterations were provided in the report). No adverse effects. Volume/record # 230/088434 contained an analysis of dosing solution. ACCEPTABLE. D. Shimer & M. Silva, 4/2/90.

012 973200 "Teratogenic Study With Orthene Technical in Albino Rats." (IBT No. B190, 9/17/71). Acephate (approximately 90%, lot SX-284) given by oral gavage at 0, 25, 100 or 200 mg/kg/day on days 6-15 of gestation, 17-21 pregnant females/group. There was a slight increase in resorption rate at the high dose, dose related decreases in maternal body weight gain, maternal toxicity considered contributory to resorption rate at 200 mg/kg and no developmental toxicity directly attributable to test article. UNACCEPTABLE (no individual animal data presented, dose level not justified, no analysis of dosing solutions, statistical analysis not provided). Not upgradeable. Purity of test article from 165, 063368, which contains the results of a 1978 audit of the raw data compared with the report. The audit found that the control data were from another study and no data were sent to the sponsor of the acephate study. Note: Initial review by J. Wong, 5/13/85 indicated a possible adverse effect. Review by D. McGee, 5-6-86, and J. Parker found no effect without maternal toxicity. Gee, 1/7/88.

EPA one-liner: teratogenic NOEL > 200 mg/kg, slightly more resorption sites per female at 200 mg/Kg than in controls, less wt. gain at 100 mg/Kg level and 200 mg/Kg by females during gestation; core grade--minimum.

161 Rebuttal of 11/20/86 to DPR review of 973200.

TERATOLOGY, RABBIT

** 146 037733 "Teratology Study in Rabbits (Technical RE 12420, Orthene)." (IRDC, 11/13/80). Acephate (92.8%, SX-1032) given by oral gavage at 0, 1, 3 or 10 mg/kg/day on days 6-27 of gestation, not adjusted for purity, with 16 per group. No significant developmental effects. Slight maternal toxicity at high dose. Developmental NOEL = >10 mg/kg, maternal NOEL = 3 mg/kg. Initially reviewed as unacceptable (McGee, 5/2/86) based on incomplete necropsy data, no analysis of dosing solutions and fetuses which aborted days 25 and 27 were not examined for malformations but study possibly upgradeable. Submission of record #061138 provides copies of records for preparation of the daily dosing solutions and 058219 - 058222 address stability in

neutral aqueous solutions. ACCEPTABLE (see Medical Toxicology Response of 6/2/88). J. Gee, 12/31/87, Davis 6/2/88.

EPA one-liner: Teratogenic NOEL => 10 mg/Kg, fetotoxic NOEL => 10 mg/Kg, maternal toxic NOEL = 3 mg/Kg; core grade--guideline.

146 037734 Positive control data for 037733 with 6-aminonicotinamide.

171 061138, 058219-058222 Stability data for 037733.

067 973196 Less complete version of record 037733. (Reviewed by J. Wong, 5/22/85, with insufficient information for evaluation.)

067 973197 Pilot study for record 037733.

161 050990 Supplement to 37733. Individual data for does #4511 and 4518.

161 Rebuttal of 11/20/86 to DPR review of 037733.

Rebuttal letter of 5/5/88. Reconsideration of all information provided led to upgrading the study to acceptable. Because acephate is quite stable under the conditions of the study, because dosing solutions were prepared daily, and because IRDC notebook pages on dosing solution preparation were provided, the lack of dosing solution analysis was not considered to invalidate the study. Davis 6/2/88.

014 973198 Invalid IBT study, 4/14/72.

GENE MUTATION

Bacterial Systems

** **101 973209** "Potential of Technical and Analytical Grade Orthene to Mutate Histidine Deficient Strains of Salmonella typhimurium (S-1202)." (Chevron, 11/28/77). Acephate (92.41%, SX-941) was tested at 0.001 to 10 mg/plate with Salmonella strain TA100 and at 1, 2 and 10 mg/plate with strains TA 98 and 1537, with and without rat liver S9. There were 2 plates/dose level. Results indicated weak mutagenic activity in TA 100 with and without activation. ACCEPTABLE by J. Wong**.** Comments by J. Gee, 9/30/86. This study included only 3 of the 4 strains listed in the guidelines. It did, however, include high amounts of acephate and repeat trials to confirm the weak effect with TA100 with the mutants per plate increasing in a concentration dependent manner but not reaching twice the spontaneous rate even at 10 mg/plate. Alone, this effect in one strain, especially TA100, would not be conclusive as to the genotoxic effect of acephate. Taken together, however, with other studies listed below, the positive effect needs evaluation. Wong, 5/20/85, Gee, 9/30/86.

EPA one-liner: Weakly positive with S. typhimurium strain TA 100 and negative with TA 98 and TA1537 strains, with or without metabolic activation; core grade--acceptable.

113 028970 "In vitro Microbiological Mutagenicity and Unscheduled DNA Synthesis Studies of Eighteen Pesticides: Excerpt for Acephate on Reverse Mutation with Salmonella typhimurium and

Escherichia coli." (SRI, 10/79). Acephate (93.5%, Lot SX-7562) was tested at 0, 1, 10, 50, 100, 500, and 1000 ug/plate (Exp. 1), 10 to 5000 ug/plate (Exp. 2), 1000 to 10,000 ug/plate (Exp. 3) and 2500 to 10,000 ug/plate (Exp. 4), with and without rat liver S9 on Salmonella strains TA98, TA100, TA1535, TA1537, and TA1538. Results indicated acephate was weakly mutagenic in TA 100. UNACCEPTABLE (only a single plating/dose level, statistical treatment of data not evident). Probably not upgradeable. Wong, 5/17/85.

EPA one-liner: Positive results on TA 98 and 100 at 5000 ug/plate and above; core grade--acceptable.

149 039417 More complete version of record 028970. Some of the objections of the initial review by J. Wong still stand [J. Gee, 9/30/86]. The data gap is filled by other studies.

113 973217 "Further Mutagenicity Studies on Pesticides (Bacterial Reversion Assay - S. typhimurium and E. coli." (Inst. Environ. Tox.-Japan, 5/18/82). Publ. in Mutation Res. 116: 185-216 (1983) -- survey of 228 pesticides in Ames test on Salmonella strains TA 98, 100, 1537 and 1538. Acephate (no purity stated) was tested at 0 to 50 mg/plate. No data - results given as "+" for TA100 and E. coli; **an increase reversion frequency above 5 mg/plate with TA100**; UNACCEPTABLE. Wong, 5/20/85.

147 973214 "Salmonella/Mammalian Microsome Mutagenicity Test (Ames Test) with six Samples of Chevron Acephate Technical and Purified (SX-911, SX-941, SX-978, SX-984, SX-986, SX-988) S. typhimurium Chevron, 12/82). Acephate (6 lots, SX-911, -941, -978, -984, -988 and -986) tested at 0 - 50 mg/plate on Salmonella strain TA100 in one trial, no activation. All lots were **weakly mutagenic in TA100.** Incomplete, UNACCEPTABLE (no metabolic activation, no repeat trials, missing data, no individual plate counts, number of platings not clear). Not upgradeable. Wong, 5/16/85, Gee, 5/12/86.

EPA one-liner: All six batches of acephate tested positive on strain TA100; core grade--supplementary.

147 016927 "Salmonella/Mammalian Microsome Mutagenicity Test (Ames Test) with Seven Samples of Chevron Acephate Technical (SX-257, SX-284, SX-357, SX-941, SX-978, SX-979 and Acetamide SX-976)." (Chevron, 12/82). Acephate (8 lots--85 to 100%, SX-257, -284, -357, -911, -941, -976, -978, -979) were tested at 0 - 50 mg/plate on Salmonella strains TA98, 100 and 1537 without activation, one trial, no individual plate counts. Seven **of 8 lots weakly positive in TA100.** UNACCEPTABLE (should TA100 read TA1537 in Table 2(?), no repeat trial, number of platings not clear, no individual plate counts, no activation included), Not upgradeable. Gee, 5/12/86.

EPA one-liner: 7 of 8 batches of acephate tested positive on strain TA 100; core grade--supplementary.

128 016927 Duplicate of 147 016927 without the analytical pages.

113 973213 Unrevised version of study identified as record 016927.

113 028970 "In vitro Microbiological Mutagenicity and Unscheduled DNA Synthesis Studies of Eighteen Pesticides: Excerpt for Acephate on Reverse Mutation with Salmonella typhimurium and Escherichia coli SRI, 10/79). Acephate (93.5%, SX-7562) tested at variable dose levels on E. coli strain WP2 +/- S9. No adverse effect indicated. UNACCEPTABLE (no description of statistical treatment of data, no individual plate counts). Possibly upgradeable. Wong, 5/17/85.

EPA one-liner: Weakly mutagenic with (5000 mg/plate) and without (6000 mg/plate) metabolic activation; core grade--acceptable.

149 039417 More complete version of record 028970. (J. Gee, 9/30/86: Some of the objections of the initial review still stand.)

Insect Systems

113 973218 "Mutagenesis Screening of Pesticides Using Drosophila Sex Linked Recessive Lethal: Chromosome Loss, Rearrangement and Nondisjunction." (WARF, 2/81). Acephate (purity not stated, no lot number) was tested at 10 ppm with 14 other pesticides in sex-linked recessive lethal assay on Drosophila melanogaster. No adverse effect reported. UNACCEPTABLE (report missing pages-including tables with acephate results). Wong, 5/17/85.

EPA one-liner: negative at 10 ppm; core grade--inadequate.

Mammalian Systems

113 973216 "Evaluation of Mutagenic Potential of Acephate Employing the L5178 TK +/- Mouse Lymphoma Assay (Forward Mutation)." (SRI, 9/80). Acephate (purity not indicated, lot SX-734 -- 93.5% in 113 973225) was tested at 10 levels between 1000-5000 ug/ml +/- rat liver S9 on mouse lymphoma cells (L5178Y). Duplicate platings/dose level with 4 hour exposure, 2-day expression time with a repeat trial. I**ncreased mutation frequency** at TK locus without S9 at 1000-5000 ug/ml and **increased mutation frequency** with S9 at 2000-5000 ug/ml. UNACCEPTABLE (need positive characterization of test article), Upgradeable. Wong, 5/17/85, Gee, 10/2/86.

EPA one-liner: Positive effects at 2000 ug/ml and above +S9 and positive effects at 1000 ug/ml and above -S9; core grade--acceptable.

** **101 973210** "L5178Y TK+/- Mouse Lymphoma Mutagenesis Assay with Chevron Acephate Technical (SX-1102)." (Microbiological Associates, 8/2/82). Acephate (technical, lot SX-1102, 98.7%) was tested at 2429, 3071, 3714, 4357 and 5000 ug/plate +/- rat liver S9 on mouse lymphoma cells (L5178Y). There were 6 platings/dose level, 4 hour exposure, 48 hr expression time. A **dose-dependent increase in mutation frequency** over entire dose range +/- S9. ACCEPTABLE. Wong, 5/21/85.

EPA one-liner: Moderately positive, with and without S9, core grade--acceptable.

** **101 973211** "Mouse Lymphoma Mutagenesis Assay with Chevron Acephate Technical (SX-762)." (Microbiological Associates, 8/2/82). Acephate (93.5%, lot SX-762) tested at 2429, 3071, 3714, 4357 and 5000 ug/plate +/- rat liver S9 on mouse lymphoma cells (L5178Y); 6 platings/dose level, 4 hour exposure and 48 hr expression time. Identical to study identified as record #97310 except a different lot of test article used. D**ose-dependent increase in mutation frequency at TK loc**us over entire dose range +/- S9. ACCEPTABLE. Wong, 5/21/85.

EPA one-liner: Moderately positive, with and without S9, core grade--acceptable.

167 058112, 058113 "Evaluation of Chevron Acephate Technical in the Mouse Somatic Cell Mutation Assay." (Hazleton, Project No. 2107-141, 10-86) Acephate technical, batch SX-1102, 98.7%, was tested in the mouse somatic cell spot test. 854 females were tested by feeding 0, 50, 200, 600 or 800 ppm acephate in the diet on days 8.5 to 12.5 of gestation, ethylnitrosourea was the positive control given at day 10.5, ip. On days 14 and 28 of lactation pups were examined for recessive coat spots. Toxic effects observed in females at 600 and 800 ppm include lacrimation, tremors, and staggered gait. The positive control was functional, no increase in recessive coat spots in acephate treated litters. UNACCEPTABLE (route of administration, no good evidence fetuses were exposed to test material) Shimer and J. Gee, 1/4/88.

Summary: Multiple reports on file with DPR contain evidence that acephate is weakly mutagenic/genotoxic in both bacterial and mammalian tests in vitro. A number of lots of acephate have been tested with Salmonella typhimurium strains with positive effects especially in strain TA100 with and without metabolic activation at high concentrations (in the mg/plate range). With mammalian cells, three reports are on file showing positive mutagenic effects in mouse lymphoma (L5178Y) in two acceptable studies and one, which is upgradeable. Three different lots were used in the mg/ml range with and without S9 activation. It should be noted that TA100 is often considered the most sensitive strain of Salmonella and L5178Y has been shown to give a higher percent of "false positives" for chemicals than, for instance, Chinese hamster cells. Some of the animal data, however, on which the evaluation of a chemical as a carcinogen/noncarcinogen is based, are not adequate, putting the "false positive" rating in some question. The fact that other test types are also positive (see below) and the reproducibility of the two tests under discussion above lend weight to the weak genotoxic effect. The in vivo mouse somatic cell mutation assay was not acceptable largely because there was no evidence presented to verify that the test article had crossed the placenta. Gee, 10/3/86 and 1/5/88.

CHROMOSOME MUTATION

112 028968, 028969 "Orthene Technical: Cholinesterase Inhibition and Cytogenetics in the Monkey, Final Report." (LSR, 1/21/83). Acephate (98.7%, lot SX-1102) was tested for SCE (028968) and chromosome aberrations (028969) at 0 and 2.5 mg/Kg only by gavage for 20 days. Peripheral

lymphocytes of monkey (Macaca fascicularis) were stimulated with phytohemagglutinin. Cells arrested in mitosis after 45 hours were incubated for 3 hours, then harvested. 1/sex/group for SCE and 1/sex/group for chromosome aberrations - lymphocytes for SCE from same animals were incubated as for aberrations but with BUDR added and incubation extended to 72 hours total. Cholinesterase inhibition was demonstrated, but no mutagenic effects noted. UNACCEPTABLE (no data included in the report), Not upgradeable. Wong, 5/21/85.

EPA one-liner: Negative at 2.5 mg/kg or body weight (only level tested) after 20 days of dosing by gavage; core grade--acceptable as supplementary.

113 973221 "Micronucleus Test on Acephate-Mice." (SRI, 3/10/80). Acephate (purity not reported but written notation of 96.6% for lot SX-734) given by gavage twice over 24 hrs at 0, 75, 150 and 300 mg/Kg to mice for micronucleus assay; justification of dose based on an oral LD50 in mice of 361 mg/kg; 24 males/group, 8 in positive control group; sampling from 8 males at 48, 72 and 96 hrs post-treatment; 500 PCE's/animal; no fatalities; no genotoxic effect reported; UNACCEPTABLE (only males tested with no justification, too few PCE's/animal, husbandry problem suggested with weight loss in controls due to "unreachable water" -- only evidence of toxicity at high dose is based on weight loss). Possibly upgradeable. Wong, 5/20/85.

EPA one-liner: Not mutagenic according to this test; core grade--minimum.

** 101 973220 "Dominant Lethal Study of Acephate Technical (SX-1102)" (Chevron, 6/11/82). Acephate (99%, lot SX-1102) given in the diet for five days at 0, 50, 500 and 1000 ppm to CD-1 mice for a dominant-lethal assay; 12 males and 190 females/group, 2 females:male for 8 weeks of mating, positive control included; no adverse effect noted. ACCEPTABLE**.** Wong, 5/21/85.

EPA one-liner: Negative, when fed to CD-1 male mice; core grade--acceptable.

012 973219 Invalid IBT study.

** 101 973212 "Cytogenetics Study in Mice Acephate Technical (SX-1102)." (E G & G Mason Res. Inst., 8-27-82) Acephate (98.7%, lot SX-1102) given by oral gavage in a single dose at 0, 11.2, 37.3 and 112 mg/Kg to Swiss white mice for a bone marrow cytogenetic assay; 4/sex/group, positive control included; clinical signs of toxicity reported; dose selection based on acute toxicity studies included with the report; no adverse effect indicated. ACCEPTABLE. Wong, 5/21/85.

EPA one-liner: Negative at 112 mg/Kg; core grade--acceptable.

**** 113 973224** "Evaluation of the Effect of Acephate on Sister Chromatid Exchange Frequencies in Cultured Chinese Hamster Ovary Cells." (SRI, 6/80). Acephate (purity not indicated, lot SX-734 with purity given as 93.5% in report 113 973225) tested at 0, 125, 250, 500, 1000 or 2000 ug/ml for 21.5 hours without S9 and at 0, 312.5, 625, 1250, 2500 or 5000 ug/ml for 2 hours with rat liver S9 activation; CHO cells in culture for SCE assay; 2 platings/dose level, positive controls included; **increase**

frequency of SCE at 500 ug/ml without S9 and at 5000 +S9. UNACCEPTABLE (test article not characterized) was the initial review by Wong. In view of the fact that the purity of this lot is contained in another report submitted at the same time, the deficiency is not considered grounds for rejecting an otherwise adequate study -- Gee. Wong, 5/20/85, Gee, 10/1/86.

EPA one-liner: Positive results without metabolic activation above 1000 ug/ml, positive results with metabolic activation at 5000 ug/ml; core grade--acceptable.

112 973222 "Mutagenicity Evaluation of Chevron Acephate Technical SX-1102 in the Sister Chromatid Exchange Assay in vivo in Mouse Bone Marrow." (Litton, 1/83). Acephate (technical, purity not stated, lot SX-1102 [purity of this lot from other reports is 99%]) given by oral gavage in a single dose at 0, 29 or 96 mg/Kg to CD-1 mice for SCE assay; 5/sex/group, positive controls included; no adverse effect indicated. UNACCEPTABLE (test article not positively characterized, inadequate number of dosing levels, report incomplete--missing appendices and tables, number of animals not indicated). Not upgradeable. Wong, 5/21/85.

EPA one-liner: Negative at 96 mg/kg; core grade--acceptable as supplementary.

158 045233 More complete version of record #973222. JGee, 9/29/86. Study is still unacceptable based on inadequate dose selection justification and lack of toxicity, no individual values and no spindle inhibitor given so inadequate number of mitotic cells were available in some groups. The reason for not evaluating slides from the 289 mg/kg group used in the preliminary study and also in the main study from dosing error is not adequate in view of the lack of m.t.d. at 96 mg/kg.

Summary: In vivo chromosome studies for dominant lethal and bone marrow chromosomal aberration formation in CD-1 and Swiss mice respectively, were both acceptable and negative for observable effects. A study for micronuclei formation in polychromatic erythrocytes, in male mice only, also showed no response to acephate but this was not an acceptable report as submitted. Another study with PHA-stimulated peripheral lymphocytes from monkeys exposed for 20 days in vivo showed no observable effect for sister chromatid exchange or chromosomal aberrations. An in vitro study with Chinese hamster ovary cells did show an increase in SCE's. This was an acceptable test. Another study on in vivo sister chromatid exchange in CD-1 mice was negative but the high dose was questionable as adequate for the test. None of the in vivo reports included good evidence that the bone marrow was exposed to a meaningful dose unlike in vitro tests where exposures of the target cells are more readily controlled. Clinical toxicity other than that to bone marrow precluded higher doses in some studies in mice (e.g., #973212). The conclusion is that there is evidence in vitro for a possible genotoxic effect. Gee, 10/3/86 and 1/5/88.

DNA DAMAGE/REPAIR

113 973225 "Differential Toxicity Assays of Nineteen Pesticides Using Salmonella typhimurium strains (DNA Damage/Repair)." (SRI, 2/81). Acephate (93.5%, SX-734) tested at 0, 1 and 5 mg/disk on Salmonella strains SL 4525 (rec+), SL 4700 (rec-), TA1978 (uvrB+) and TA1538 (uvrB-) in a spot test

for differential toxicity without metabolic activation; 2 platings/dose level, positive controls included; two trials; no adverse effects reported in first trial but **differential growth** reported with SL (rec) strains in second trial: rec+ with 9 mm and rec- with 12 mm zone of inhibition (6mm disk); UNACCEPTABLE (no activation included). Review by Gee identifies a possible adverse effect. Wong, 5/20/85, Gee, 10/1/86.

EPA one-liner: Negative up to 5 mg; core grade--acceptable.

113 028972 "In vitro Microbiological Mutagenicity and Unscheduled DNA Synthesis Studies of Eighteen Pesticides: Excerpt for Acephate on Differential Toxicology in Escherichia coli and Bacillus subtilis." (SRI, 10-79) Acephate (93.5%,, lot SC-7562) tested at 0.01, 0.10, 1.0 and 5.0 mg/disc/plate on E. coli strain W3110/p3478 and B. subtilis strains H17/M45 in a spot test (damage/repair); 1 plate/dose level, no repeat trial; no adverse effect indicated. UNACCEPTABLE (single plating and no repeat trial, no activation.) Reason why B. subtilis

H17/M45 (rec +/-) did not show differential effect as did Salmonella (#973225) is not clear. Wong, 5/17/85.

EPA one-liner: negative; core grade -- unacceptable.

149 039419 More complete version of record 028972. Gee, 10/1/86. The objections in the initial review stand.

113 028971 "In vitro Microbiological Mutagenicity and Unscheduled DNA Synthesis Studies of Eighteen Pesticides: Excerpt for Acephate on Mitotic Recombination with Saccharomyces cerevisiae." (SRI, 10/79). Acephate (93.5%, lot SX-7562) tested at 0, 0.1, 0.5, 1.0 and 5.0 % (trial 1) and at 1, 2, 4 or 5% (trial 2) +/-S9 on S. cerevisiae strain D3 in a mitotic recombination assay; incubated for 4 hours on a roller drum, then diluted serially and plated on 5 plates for survivors x 10^{-5} and 3 plates for mitotic recombinants x 10^{-3}; **positive effects at 1%** and above with and without metabolic activation. UNACCEPTABLE (dose selection not justified with marginal cytotoxicity demonstrated, no individual plate counts and no statistical analysis reported, use of DMSO as solvent is not recommended.) Wong, 5/17/85.

EPA one-liner: Positive at 1% and above; core grade--acceptable.

149 039418 More complete version of record 028971. Gee, 10/1/86. Evaluation stands.

113 973215 "Orthene Technical: Cholinesterase Inhibition and Mitotic Gene Mutation, and Reverse Mutation with S. cerevisiae D7 for 7 Pesticides - Orthene." (SRI, 6/80). Acephate (93.5%, lot SX-734) tested at 0, 1, 2, 3, 4 and 5% +/- rat liver S9 on S. cerevisiae strain D7 (diploid) in mitotic crossing over and gene conversion assays; repeat test using 3, 3.5, 4, 4.5 and 5%; incubated for 4 hours, then diluted and plated; with S9, **an increase in mitotic crossing over and reverse mutation at 2% and above-- increased frequency of gene conversion at 1% and above; without S9, an increase in frequency of crossing over, reverse mutation and gene conversion** at 1% and above.

UNACCEPTABLE (number of plates/group not clear, no rationale for dosing levels, individual plate data not included, methods of statistical treatment not clear), Possibly upgradeable. Wong, 5/16/85.

EPA one-liner: Positive for crossing over , gene conversion and reverse mutation at 1% and above without metabolic activation, positive for gene conversion at 1% and above, positive for crossing over and reverse mutation at 2% and above with metabolic activation; core grade--acceptable.

**** 113 028973** "In vitro Microbiological Mutagenicity and Unscheduled DNA Synthesis Studies of Eighteen Pesticides: Excerpt for Acephate on Unscheduled DNA Synthesis." (SRI, 10/79). Acephate (93.5%, lot SX-734) tested at 0.1 to 72 ug/ml without S9 (Exp. #1), 125 to 2000 ug/ml without S9 (Exp. #2), 0.1 to 1000 ug/ml with rat liver S9 (Exp. #3) and 250 to 4000 ug/ml (Exp. #4) with contact-inhibited WI-38 human fibroblasts in UDS assay; 3 hour exposure without activation and 1 hour with activation; hydroxyurea to block semiconservative DNA synthesis; DNA was extracted, DNA determined by diphenylamine reaction and tritium quantitated by liquid scintillation counting; in the absence of activation, slight increase in UDS at and above 1000 ug/ml. Initial review by Wong indicated an incomplete report with no protocol submitted. 149 039428 contains

the full document making the study ACCEPTABLE with **an adverse, genotoxic effect** (Gee, 10/1/86). Wong, 5/17/85,

EPA one-liner: Positive response without metabolic activation at 1000 ug/ml and above; core grade--acceptable.

149 039420 More complete version of record 028973. Gee, 10/1/86. See above.

Summary: Comparison of differential toxicity in repair proficient versus repair deficient strains of Salmonella suggest an adverse effect on viability of cells with a defective recombinant repair pathway (rec-), while the UV-repair deficient strain (uvrB-) grew approximately the same as the uvrB+ strain. Bacillus subtilis rec+/- strains, however, did not show any difference in growth for reasons that are not known. On the other hand, Saccharomyces cerevisiae D3 and D7 both showed increased mitotic recombination, mitotic crossing-over and gene conversion with exposure to acephate, lending support to the data with Salmonella. In these tests, DNA damage occurs of a type, which is repaired by DNA recombination. When a cell cannot perform this function, it is killed reproductively. In proficient strains, repair occurs, allowing for survival or, in Saccharomyces, enhancing mitotic crossing over, which is essentially a test of repair. In addition, there was a slight increase in unscheduled DNA synthesis in mammalian cells, substantiating the results in microbial systems. Gee, 10/3/86 and 1/5/88.

SUMMARY OF GENOTOXICITY STUDIES: Taken altogether, the studies in the three areas indicate the in vitro tests reported to DPR were more sensitive than the in vivo genotoxicity studies submitted or acephate is nonmutagenic in vivo. The possibility of in vivo effects should not, however, be dismissed 1) because correlation of in vitro to in vivo effects is not well understood and 2) in vivo tests in other areas on file suggest adverse oncogenic effects. Full assessment of these effects cannot be made unless adequate in vivo studies in the area of genotoxicity are available. The data requirements are fulfilled by the in vitro studies. Gee, 10/3/86 and 1/5/88.

NEUROTOXICITY

** 151 039603, 039602 "Acute Delayed Neurotoxic Study in Chickens with Chevron Acephate Technical Final Report and Addendum." (Wildlife International, 10/18/85). Acephate (98%) at 785 mg/Kg, redosed after 21 days, 5 mg/kg atropine to protect at dosing with additional atropine given over 21 hours; 6 hens in control groups and 12 in treatment group; TOPC positive control; no delayed neurotoxicity note. ACCEPTABLE. McGee, 4/21/86.

** 067 973171 "Studies on Acute Delayed Neurotoxicity of Orthene (Chickens)." (Bozo Research Center-Japan, 11/79). Acephate (98.9%) at 375 mg/Kg by gavage, one dosing, 5 mg/kg atropine to protect; 12 hens in control groups and 24 in treatment group; TOPC positive control; no delayed neurotoxicity noted. ACCEPTABLE**.** Wong, 5/22/85.

EPA one-liner: Negative, but insufficient; core grade-supplementary.

015 973172 Invalid IBT study, 1/20/72.

**** 108-286 153408** " Subchronic (13-Week) Neurotoxicity Study of ORTHENE Technical in Rats." (M.D. Nemec; WIL Research Laboratories, Inc., Ashland, OH; Project ID. No. WIL-194014; 1/16/97)
Thirty Sprague-Dawley rats/sex/group were dosed orally in the diet with 0, 5, 50 or 700 ppm of ORTHENE Technical (lot no. SX1725, purity: 99.0%) for up to 13 weeks ((M): 0, 0.33, 3.31, 48.63 mg/kg/day, (F) 0, 0.41, 3.95, 58.27 mg/kg/day). There were no treatment-related effects upon mean body weights or food consumption. Although some of the FOB parameters and motor activity measurements for the treated animals were statistically different from that of the control group, there was no consistent pattern of effect noted over the course of the study. The mean cholinesterase (ChE) activity levels for plasma were lower than that of the control at 3 weeks for the 50 ppm males and females ($p<0.05$ or $p<0.01$) and at 3, 7 and 13 weeks for the 700 ppm males and females ($p<0.01$). The mean red blood cell activity levels were less at 3, 7 and 13 weeks for the 700 ppm males ($p<0.01$) and at 3 and 7 weeks for the 700 ppm females ($p<0.05$ or $p<0.01$). In the 6 subregions of the brain for which ChE activity was assayed, the activity levels were less than that of the control at 50 ppm and above for all of the regions at least twice for the 3 time points assayed ($p<0.1$). The mean percent of control activity for the 50 ppm males ranged from 50.6% in the cortex (week 13) to 75.8% in the cerebellum (week 13). For the 50 ppm females, the percent of control activity ranged from 45.4% in the hippocampus (week 13) to 81.5% in the brainstem (week 3). For the 5 ppm treatment group males , ChE activity was reduced in the hippocampus (week 3, $p<0.01$, 86.1% of control), midbrain (weeks 3, 7,13, $p<0.01$, 85.5 to 90.5%), brainstem (weeks 3, 7, 13, $p<0.01$, 85.0 to 90.6%), cerebellum (week 3, $p<0.01$, 89.1%), and cortex (weeks 3, 7, 13, $p<0.01$, 82.4 to 89.9%). Similarly, for the 5 ppm females, ChE activity was less in the hippocampus (week 13, $p<0.01$, 71.6% of control), olfactory lobe (weeks 7, 13, $p<0.05$, 75.7, 82.2%), midbrain (weeks 3, 7, 13, $p<0.1$ or $p<0.05$, 80.9 to 91.0%), cerebellum (week 7, $p<0.1$, 83.9%) and cortex (week 13, $p<0.01$, 86.2%). No treatment-related effects were noted in the histopathological examination. **Possible adverse effect:** significant brain ChE inhibition; **NOEL (Clinical Signs):** (M/F) 700 ppm ((M): 48.63 mg/kg/day, (F):

58.27 mg/kg/day) (based upon the lack of treatment effects in the FOB determinations for the 700 ppm group); **NOEL (ChE Inhibition):** < 5 ppm ((M): < 0.33 mg/kg/day, (F) < 0.41 mg/kg/day) (based upon significant brain ChE inhibition at 5 ppm). **Study acceptable.** (Moore, 2/6/02)

**** 108-285 153407** "An Acute Neurotoxicity Study of Orthene® Technical in Rats" (M.D. Nemec; WIL Research Laboratories, Inc., Ashland, OH; Project ID No. WIL-194013; 12/9/96.) Thirty Sprague-Dawley rats/sex/group were dosed by oral gavage with 0, 10, 100 or 500 mg/kg of Orthene Technical (lot no. SX1725; purity: 99.0%). Twelve animals/sex/group were evaluated in the Functional Observational Battery (FOB) and for locomotor activity as well as in the neuropathology examination. The other 18 animals/sex/group were euthanized for cholinesterase (ChE) activity evaluation, 6 animals/sex/group at 2.5 hours and 7 and 14 days post-dose. No animals died as a result of the treatment. The mean body weight for the males in the 500 mg/kg group was less than that of the controls at 7 days post-dose ($p<0.05$). Clinical signs included whole body tremors, repetitive movement of the mouth, tremors of the forelimbs and/or hindlimbs, and alterations in the posture/gait of the animals in the 100 and 500 mg/kg groups in a dose-related manner. The earliest observation of these signs was at 30 minutes and the time to peak effect was between 2 and 2.5 hours and persisted up to 8 hours post-dose in the high dose group. The signs were no longer present by the next day. Salivation, lacrimation and chromodaccryorrhea was observed only in the 500 mg/kg animals. In the FOB, at 2.5 hours post-dose, the 100 and 500 mg/kg animals exhibited in a dose-related manner, abnormal posture, whole body tremors which ranged from slight to extremely coarse, slightly impaired to totally impaired mobility, walking on tiptoes to ataxia, decreased arousal and rearing, diminished response to a tail pinch, impaired righting reflex, and reduced body temperature. In addition, the high dose animals exhibited signs of lacrimation, salivation, poor grooming, diminished startle, touch and approach responses and catalepsy. The males in the high dose group had no pupilary response. The hindlimb extensor strength was reduce for the males in both the 100 and 500 mg/kg groups and the females in the 500 mg/kg group. The fore and hindlimb grip strength for the high dose males was reduced from that of the controls ($p<0.01$ and $p<0.05$, respectively). The rotorod performance was affected in all of the male treatment groups ($p<0.01$) and for the high dose females ($p<0.01$). In the motor activity evaluation, total activity and ambulatory activity counts were reduced for the animals in the 100 and 500 mg/kg groups. All of these parameters had returned to normal by day 7. In the ChE activity determinations, all of the treatment groups for both sexes exhibited reduced activity levels in the plasma, red blood cells and subregions of the brain at 2.5 hours post-dose ($p<0.01$). At 7 days post-dose, reduced activity was still evident in the following tissues: hippocampus, females, 500 mg/kg; midbrain, males, 500 mg/kg, females, 500 mg/kg; brainstem, males and females, 500 mg/kg; cerebellum, males and females, 500 mg/kg; cortex, males, 500 mg/kg, females, 10, 100, 500 mg/kg ($p<0.05$ or $p<0.01$). At 14 days, activity was reduced in the red blood cells and the midbrain of the 500 mg/kg males ($p<0.05$ and $p<0.01$, respectively). No treatment related lesions were evident in the neuropathology examination. **Possible Adverse effect:** extensive neurtoxic signs; **NOEL(clinical signs) (M/F):** 10 mg/kg (based upon the clinical signs manifested by the 100 mg/kg treatment animals); **NOEL (cholinesterase inhibition):** < 10 mg/kg (based upon ChE inhibition evident in the plasma, red blood cells and subregions of the brain of the 10 mg/kg treatment group); **Study acceptable.** (Moore, 10/16/01)

108-284 153406 " Range-Finding Acute Study of Orthene® Technical in Rats" (M.D. Nemec; WIL Research Laboratories, Inc., Ashland, OH; Project No. WIL-194015; 4/27/95) Two Sprague-Dawley

rats/sex/group were dosed by oral gavage with 0, 5, 25, 125 or 500 mg/kg of Orthene® Technical (batch no. SX 1725, purity: 99.4%) in Phase I. In Phase II, five females/group were dosed orally with 0, 0.5, 2.5 or 5 mg/kg of the test material. In Phase I, animals received detailed clinical examinations at 15 and 30 minutes and 1, 2 and 2.5 hours post-dose. In Phase II, the animals were examined at the time of euthanization, 2.5 hours post-dose. Plasma, red blood cell and sections of the brain were assayed for cholinesterase (ChE) activity at the time of when peak toxic signs were manifested, 2.5 hours post-dose. No animals died as a result of the treatment. Clinical signs of repetitive mouth movements, tremors in the forelimbs/hindlimbs or whole body, salivation and altered gait were noted in the 125 and 500 mg/kg groups. One male in the 25 mg/kg group exhibited repetitive mouth movements. Twitching of both ears was noted for animals in the 25, 125 and 500 mg/kg treatment groups. Hypothermia was exhibited by the animals in the 500 mg/kg. In Phase I, cholinesterase activity was reduced in a dose-related manner with the activity level ranging from 68.9 to 80.5% and 69.6 to 85.9% of the control activity for the males and females, respectively, in the 5 mg/kg treatment group. In Phase II, at 2.5 mg/kg, female ChE activity in the various brain sections ranged from 78.4 (brain stem) to 86.6% (hippocampus) of the control activity. At 0.5 mg/kg, the activity levels in the brain ranged from 92.1 to 102.8% of control activity. **Possible adverse effects**: signs of neurotoxicity. **NOEL (ChE inhibition): (M)** < 5 mg/kg (based upon the brain ChE inhibition exhibited by the males in the 5 mg/kg group), **(F)** 0.5 mg/kg (based upon the inhibition of ChE in the brain of the females in the 2.5 mg/kg group); **NOEL (clinical signs): (M/F)** 5 mg/kg (based upon the signs observed in the 25 mg/kg animals). **Study supplemental.** (Moore, 10/19/01)

108-315 183892 " A Single Oral Dose Study with Acephate Technical in Humans" (S. Freestone and P. McFarlane; Inveresk Research, Elphinstone Research Centre, Tranent, EH33 2NE, Scotland; Project ID. ICR 013072; 5/3/00, 1st Amend. 6/26/00, 2nd Amend. 3/23/01) Four groups of 10 male subjects each and one group of 10 female subjects were included in the study. For each group, 7 people were dosed with the test material and 3 received the lactose placebo. The male groups were treated with one dose in a gelatin capsule of 0.35, 0.7, 1.0 or 1.25 mg/kg of Acephate Technical (lot no. 80121 (SCC), purity: 99.0%). The female group received 1.0 mg/kg of the test material. Each subject was screened prior to treatment in which hematology, clinical chemistry, vital signs and ECG were evaluated. Plasma and red blood cell (RBC) cholinesterase (ChE) activities were measured 6 times prior to dosing with the mean values being used as the base line. During the study, blood samples were recovered at 1, 2, 4, 8, 12, 24, 48 and 72 hours and 7 and 14 days post-dose. ChE activity measurements and analysis of acephate and methamidophos levels were performed. Hematology, clinical chemistry and urinalysis parameters were evaluated at 24 hours post-dose. Vital signs and ECG were measured at 2, 4, 8, and 24 hours post-dose. No treatment-related effects were noted for the vital signs, ECG, hematology, clinical chemistry and urinalysis. Analysis of the ChE activity data revealed a statistically significant % change from baseline for plasma ChE at 12 (-12.77%, $p<0.01$), 24 (-8.89%, $p<0.01$) and 48 (-9.12, $p<0.001$) hours post-dose for the males in the 1.25 mg/kg group and at 8 (12.73%, $p<0.05$), 12 (-12.08%, $p<0.05$) and 24 (-10.50%, $p<0.01$) hours for the females in the 1.0 mg/kg group. For RBC ChE, a statistically significant % change from baseline was noted for the males at 12 hours post-dose (-6.75%, $p<0.01$). In the pharmacokinetic analysis, the $T_{1/2}$ elimination ranged from 4.39 to 5.42 hours. The time to maximal concentration in the blood (T_{max}) ranged from 1.29 to 2.71 hours. The highest mean concentration of both acephate and methamidophos recovered in the urine occurred during the 0 to 12 hours post-dose interval. For the males, the mean percentage of the administered dose recovered in the urine up to 48 hours post-

dose ranged from 43.3% for the 0.35 mg/kg group to 52.5% for the 1.0 mg/kg group. The mean percentage of the administered dose recovered in the urine from 0 to 48 hours post-dose from the 1.0 mg/kg females was 26.0%. **No adverse effects indicated. NOEL: (M)** 1.0 mg/kg (based upon a statistically significant reduction in the plasma ChE activity for the 1.25 mg/kg males); **(F)** < 1.0 mg/kg (based upon a statistically significant reduction in plasma ChE activity for the 1.0 mg/kg females). **Study supplemental.** (Moore, 12/4/01)

108-200; 67747; AThe Cholinesterase Inhibition Potential of Acephate Techical (SX-1102) Following 4-, 9-, or 13-Week Dietary Administration in Male and Female Rats@; (G.P. Brorby, and D.W. Rosenberg; Chevron Environmental Health Center, Inc., Richmond, CA; Study No. S-3068; 12/30/87); Thirty Sprague-Dawley rats/sex/group were treated in the diet with 0, 2, 5, 10 or 150 ppm of Orthene Technical (lot no. SX-110,; purity: 98.2%) for up to 13 weeks ((M), 0, 0.12, 0.28, 0.58, or 8.90 mg/kg/day, (F) 0, 0.15, 0.36, 0.76 or 11.48 mg/kg/day). Ten animals/sex/group were euthanized after 4, 9 and 13 weeks on study. There were no mortalities during the study. There were no treatment-related clinical signs or effects on food consumption. There was no apparent treatment-related effect on body weight gain. Significant brain cholinesterase (ChE) inhibition was noted for the 5 ppm group and above for both sexes after 4, 9 and 13 weeks of treatment (p<0.01) (% of control activity: 5 (M) 92.5 to 92.7%, (F) 89.3 to 91.2%, 10 (M) 85.8 to 89.1%, (F) 83.1 to 88.5%, 150 (M) 48.3 to 52.7%, (F) 45.2 to 54.0%). For the 2 ppm treatment group, only the females demonstrated significant brain ChE inhibition at all of the time points (p<0.01) (% of control activity: 90.3 to 91.9%). A dose-response for ChE inhibition in the plasma and red blood cells was not well demonstrated with statistical significance only at the 150 ppm treatment level. The necropsy examination did not reveal any treatment-related lesions. **Possible adverse effect:** inhibition of brain cholinesterase; **NOEL:** (M) 2 ppm (0.12 mg/kg/day) (based upon significant brain ChE inhibition in the 5 ppm treatment group, (F) < 2 ppm (< 0.15 mg/kg/day) (based on the significant 108-200; 67747; AThe Cholinesterase Inhibition Potential of Acephate Techical (SX-1102) Following 4-, 9-, or 13-Week Dietary Administration in Male and Female Rats@; (G.P. Brorby, and D.W. Rosenberg; Chevron Environmental Health Center, Inc., Richmond, CA; Study No. S-3068; 12/30/87); Thirty Sprague-Dawley rats/sex/group were treated in the diet with 0, 2, 5, 10 or 150 ppm of Orthene Technical (lot no. SX-110,; purity: 98.2%) for up to 13 weeks ((M), 0, 0.12, 0.28, 0.58, or 8.90 mg/kg/day, (F) 0, 0.15, 0.36, 0.76 or 11.48 mg/kg/day). Ten animals/sex/group were euthanized after 4, 9 and 13 weeks on study. There were no mortalities during the study. There were no treatment-related clinical signs or effects on food consumption. There was no apparent treatment-related effect on body weight gain. Significant brain cholinesterase (ChE) inhibition was noted for the 5 ppm group and above for both sexes after 4, 9 and 13 weeks of treatment (p<0.01) (% of control activity: 5 (M) 92.5 to 92.7%, (F) 89.3 to 91.2%, 10 (M) 85.8 to 89.1%, (F) 83.1 to 88.5%, 150 (M) 48.3 to 52.7%, (F) 45.2 to 54.0%). For the 2 ppm treatment group, only the females demonstrated significant brain ChE inhibition at all of the time points (p<0.01) (% of control activity: 90.3 to 91.9%). A dose-response for ChE inhibition in the plasma and red blood cells was not well demonstrated with statistical significance only at the 150 ppm treatment level. The necropsy examination did not reveal any treatment-related lesions. **Possible adverse effect:** inhibition of brain cholinesterase; **NOEL:** (M) 2 ppm (0.12 mg/kg/day) (based upon significant brain ChE inhibition in the 5 ppm treatment group, (F) < 2 ppm (< 0.15 mg/kg/day) (based on the significant brain ChE inhibition in the 2 ppm treatment group). **Study acceptable.** (Moore, 2/27/02)

MISCELLANEOUS

Guidance for the Reregistration of Pesticide Products Containing Acephate as the Active Ingredient, EPA, September, 1987, gives the following data gaps for acephate: Twenty-one day inhalation study in rats, chronic toxicity in the rat to determine the NOEL for brain cholinesterase inhibition and rat reproduction

study to establish the NOEL - this has been satisfied with DPR Record #060979, not included in the EPA review. Acephate has been classified as a class C carcinogen or "possible" human carcinogen based on the increase in liver adenomas/carcinomas and hyperplastic nodules in female mice only at the high dose at term plus the positive findings in in vitro mutagenicity tests. In vivo studies were negative for genotoxicity.

Methamidophos: Technical acephate contains 0.9 to 1.2 % w/w methamidophos*, a cholinesterase inhibitor and a metabolite of acephate as well as a contaminant. By acute studies, it is highly toxic, being category I. Methamidophos was not oncogenic at 25 ppm in rats and not teratogenic in rabbits (2.5 mg/kg) or in rats (3.0 mg/kg). In a 1-year dog study and a 2-year rat study, inhibition of brain cholinesterase was observed at 2 ppm (0.05 mg/kg/day) (LDT). The EPA reregistration document identifies a rat reproduction study and mutagenicity studies as remaining data gaps.

Methylthioacetate: This is an impurity* in the currently registered product. According to EPA, additional studies (acutes and 90-day dermal in rabbits) are required. Also, they indicate that a battery of mutagenicity tests in addition to the positive mouse lymphoma test are needed.

*Chevron's rebuttal letter of 5/5/88 states that current manufacturing processes produce > 99.9% pure acephate.

Twelve studies on methylthioacetate to support continued registration of acephate were submitted by Valent U.S.A. Corp. with a letter dated 12/5/88. The studies include acute, subchronic and mutagenicity. These studies are under tolerance number 51656. One-liners created in the 950 review follow.

108-128 16926 Review of the results of mutagenicity testing of Acephate technical (Orthene): Gene Mutation, Primary DNA Damage, Chromosome Alteration. No worksheet. Kellner, 5/5/93.

SUBCHRONIC, DERMAL

51656-003 72296 "Ninety Day Dermal Toxicity Study in Rabbits with Methylthioacetate (MTA)" (Chevron Environmental Health Center, Inc. No. CEHC 2822, 1-15-88) Methylthioacetate, SX-1732, 98.9%, was placed on the backs of New Zealand White rabbits for 6 hours/day, 5 days/week at dose levels of 0, 5, 20 or 60 mg/kg, deaths by percent were 7, 50, 53 and 50, respectively. There were 15/sex/group initially except for the low dose group which had 10/sex. The majority of deaths were attributed to mucoid enteritis. Clinical signs include inappetence, diarrhea, no stool and decreased activity related to mucoid enteritis. At 20 and 60 mg/kg severe skin irritation occurred until the site of application was varied on the animal. Histopathology revealed no compound related lesions of the optic nerve or liver, the known target organs in acute studies. Supplemental since not an active pesticidal ingredient, otherwise UNACCEPTABLE, and not upgradeable. The occurrence of disease compromised the value of the study and reduced animal numbers to unacceptable levels. D. Shimer, 7/13/89.

GENE MUTATION

51656-004 072297 "Salmonella/Mammalian Microsome Plate Incorporation Mutagenicity Assay (Ames Test) with Methylthioacetate (SX-1732)," (Microbiological Associates, Study No. T5771.501014, 12-23-87) Methylthioacetate (98.2% pure; LOT #: SX-1732), was tested for mutagenicity with Salmonella typhimurium strains TA98, TA100, TA1535, TA1537 and TA1538 at concentrations of 10,000, 5000, 2500, 500 or 100 ug/plate (triplicate plates). Assays were done in the presence and absence of metabolic activation. A confirming repeat test was performed. No increase in the number of revertants was observed. No adverse effects. Supplemental**.** This is not a registered pesticide, but a contaminant present at a very low concentration in Acephate. D. Shimer & M. Silva, 3/30/90.

CHROMOSOME MUTATION

51656-004 072298 "Clastogenic Evaluation of Methylthioacetate (SX-1732) in the Rat Bone Marrow Cytogenetic Assay Following a Four-Day Inhalation Pilot Study," (Hazleton Laboratories America, Inc., Study No. 2107-147, 11-12-87). Fischer 344 rats, 5/sex/group, were exposed to methylthioacetate (Lot #: SX-1732; 98.2% pure) vapors at concentrations of 400, 600 or 800 ppm for 6 hours/day for 4 consecutive days. Animals were sacrificed 19 hours after their last exposure and 50 cells/dose/animal were scored (50 spreads/animal). NOEL < 400 ppm (Clinical signs were observed at ≥ 400 ppm; body weights in both sexes were significantly decreased at ≥ 400 ppm; there was a significant decrease in food consumption at ≥ 400 ppm; 80% mortality--all males and 3/5 females--at 800 ppm). NOAEL = 600 ppm. No positive control, was used in this study, however, the high dose was acceptable, due to the degree of mortality at 800 ppm. No adverse effects. Supplemental. The study was considered to be a pilot study. This is not a registered active ingredient, but a contaminant in Acephate. D. Shimer & M. Silva, 3/29/90.

DNA DAMAGE/REPAIR

51656-004 072299 "Micronucleus Assay in Mouse Bone Marrow Erythrocytes Following Inhalation Exposure to Methylthioacetate (SX-1763, 99.2% Purity)," (Chevron Environmental Health Center, Inc., No. CEHC 2751, 1-15-88). Methylthioacetate (Lot #: SX-1763; 99.2% pure) was used, as a vapor, on Swiss albino mice (≥ 15 mice/sex/group) for 4 hours at actual concentrations of 0, 445, 651 or 796 ppm. Five/sex/group were sacrificed at 24, 48 and 72 hours after the start of exposure. 1000 PCE were examined/slide/animal, 1000 cells were counted to determine NCE:PCE ratio. No adverse chromosome effects. No treatment related increase in the number of micronucleus was observed, however, 5 animals died at 796 ppm and histopathology revealed treatment related lesions in lungs of all methylthioacetate dosed animals. Supplemental. This is not a registered ingredient, but has been submitted because it is a contaminant in Acephate. D. Shimer & M. Silva,